30

CLAIMS

Process for the production of a biological substance by perfusion culturing of suspended animal cells in a serum free cell culture medium, wherein the biological substance is separated from the cells by filtration, characterized in that at least 0.001 w/w% of polyoxyalkylene sorbitan fatty acid ester represented by formula 1,

$$R^{1}O(AO)_{n}$$
 $O(AO)_{o}R^{2}$
 $CH_{2}O(AO)_{p}R^{3}$
 $O(AO)_{q}R^{4}$
(1)

- wherein R¹, R², R³ and R⁴ each independently represent H or a fatty acid restgroup, i.e. the remains of a condensation of a fatty acid and an alcohol, provided that at least one of R¹ through R⁴ is a fatty acid restgroup, wherein A represents an ethylene or propylene group and n, o, p and q each independently represent values from 0 to 100, is present in the cell culture medium.
 - Process according to claim 1, characterized in that in formula 1, the sum of n, o, p, and q is from 50 to 300.
 - 3. Process according to claim 1 or claim 2, characterized in that at least 0.01 w/w% of the compound of formula 1 is present in the cell culture medium.
- 20 4. Process according to any of claims 1-3, characterized in that the animal cells are mammalian cells.
 - Process according to any of claims 1-4, characterized in that the compound of formula 1 is a Tween[™] compound.
- 6. Process according to any of claims 1-5, characterized in that the filtration is performed with an internal filter.
 - 7. Process according to claim 6, characterized in that the internal filter is a spinfilter.
 - 8. Process according to any of claims 1-7, characterized in that the biological substance is a biopharmaceutical product.

WO 2004/097006 PCT/NL2004/000300 - 8 -

- 9. Process according to claim 8, characterized in that the serum free cell culture medium is also a mammalian source free medium.
- 10. Process according to any of claims 1-9, characterized in that the biological substance is further purified by downstream processing.

5